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Short communication

FIRST REPORT OF ROOT ROT CAUSED BY *PYTHIUM APHANIDERMATUM* AND *PYTHIUM* F-GROUP ON HYDROPONICALLY GROWN PEPPER IN BULGARIA

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Abstract

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In September 2013 and July 2014, severe wilt symptoms were observed on mature, sweet pepper (*Capsicum annum* L.) plants (c. Sofijska kapiya) grown in a rockwool hydroponic system in a large-scale commercial greenhouse near Petrich, South West of Bulgaria. Examination of the wilted plants revealed progressive chlorosis of the lower leaves, severe brown and soft root rot and basal stem rot. Less affected plants showed yellow discoloration of the root system, stunted growth and lack of vigor. Approximately 25% of the pepper plants collapsed and died particularly during the fruiting period in the first year of observations and more than 30% of plants died in the second year. Isolations were made from sections of roots and basal stems of pepper plants with symptoms of disease on standard, nonselective media such as oatmeal (OA), potato dextrose (PDA) or water (WA) agar media. Plates were incubated at 25°C in the dark for 7 days. *Pythium* spp. isolates were readily obtained from all pepper plants with disease symptoms as well as from symptomless plant roots.

Twelve representative colonies were selected and purified by subculturing on OA medium. Observations on colony morphology and growth were made on WA, OA and cornmeal agar (CMA) at 25°C. Colonies were fast growing, without a special pattern, with white, cottony aerial mycelium, somewhat loose on WA, and with fluffy aerial mycelia on OA and CMA. Three isolates (ISSAPP deposit numbers PEP65B1, PEP66A2 and PEP68D1) were identified as *Pythium aphanidermatum* (Edson) Fitzp., using a grass leaf culture technique as described by Waterhouse (1967). Microscopic characteristics of these isolates included branched, aseptate, hyaline hyphae, up to 11 µm wide. Sporangia were mostly terminal or occasionally intercalary, consisting of swollen, torulated hyphal branches. Zoospores formed at room temperature. Oogonia and antheridia formed abundantly both on agar media and in water cultures. Oogonia were terminal, globose and smooth, 15.8 to 31.5 µm in diameter. Antheridia were cylindrical to rotund, either terminal or intercalary, diclinous or occasionally monoclinous, one or two per oogonium, 10.2 to 17.4 µm in diameter. Antheridial cells were broadly attached to the oogonium. Oospores were globose, smooth and aplerotic, 12.3 to 26.3 µm in diameter. The rest nine isolates obtained from pepper root system formed noninflated, filamentous sporangia, did not produce sexual reproductive organs in single or dual cultures and thus were designated as *Pythium* "Group F" (*Pythium* sp. F) according to Waterhouse (1968) and van der Plaats-Niterink (1981). Pathogenicity of each of these isolates was tested twice. Pepper seedlings (c. Sofijska kapiya) were grown individually in 10 x 10 cm rockwool cubes saturated

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with nutrient solution (pH *ca*. 6.5, electrical conductivity of 2.3–2.4 $mS.cm^{-1}$, Rosella Ltd., Sofia, Bulgaria) until three to four true leaves formed. The growing cubes were placed on standard rockwool slabs (130 × 20 × 10 cm) arranged on a concrete floor in experimental glasshouse and preliminarily saturated with the nutrient solution. *Pythium* isolates were cultivated in 15 cm Petri plates containing 20 ml of Oatmeal broth. Plates were incubated for 14 days at 26°C in the dark. Mycelial mats of each isolate were collected, gently washed under running water to remove medium residues, mixed and blended at high speed in a blender. The inoculum of each isolate was suspended in sterile distilled water (*ca*. 10^6 cfu ml^{-1}) and 100 ml of this suspension was added uniformly on the surface of a single rockwool cube around the stem base of the seedling plant. Each slab contained nine cucumber plants inoculated with the same *Pythium* isolate. Negative controls were pepper seedlings inoculated similarly with equal amount of sterile distilled water. Inoculated plants were kept for 26 to 50 days in experimental glasshouse at 18-32°C and monitored every three days for appearance of disease symptoms. Symptoms, very similar to those expressed in the original host-plants, followed by plant mortality were developed on up to 66.7% of the seedlings artificially inoculated with the respective *Pythium* isolate. The inoculated pathogens were consistently reisolated from symptomatic tissues, thereby fulfilling Koch's postulates. None of the control plants developed symptoms of disease. Pythium root rot on hydroponically-grown peppers and many other crop species has been reviewed by Sutton et al. (2006). *Pythium aphanidermatum* has been described on peppers in Bulgaria elsewhere (Vanev et al., 1993). To our knowledge, this is the first report of the pathogen on hydroponically grown pepper in the country and the first report of *Pythium* F-group in Bulgaria.

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